

A CONFORMATIONAL MODEL OF SERINE TRANSFER RNA PROPOSED ON THE BASIS OF ELECTRON MICROSCOPY

L.O.FRÖHOLM and B.R.OLSEN

*Methodology Department, National Institute of Public Health, Oslo, and
Anatomical Institute, University of Oslo, Norway*

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Based on the cloverleaf arrangement of Holley et al. [1] several three dimensional structures have been proposed for tRNA molecules in solution [2–4] and a detailed model of the anticodon arm has been built by Fuller and Hodgson [2]. Guschlbauer [6] proposed models based on triple-strand arrangements.

Results recently obtained by electron microscopy [7, 8] indicated that we would have to modify the currently available models in order to make them fit the observed structures. Thus positive staining of tRNA with uranyl acetate showed the presence of thread-like structures 80–100 Å in length and 20 Å in diameter. In addition to large net-like aggregates composed of these elements, long threads, several hundred Å in length, were observed. The latter can conceivably be explained by assuming end-to-end polymerization of the short elements. Sometimes the 80–100 Å particles formed V-shaped structures. End-to-end complexes of these could explain the rhombic structures with sides 50 Å in length reported before [7,8]. Occasionally the short elements displayed a segmented substructure.

In recent experiments with a tRNA^{Ser} fraction [9], (obtained through the courtesy of H.G.Zachau) which on Sephadex behaves as dimers, short rods of high contrast were observed after positive staining (see fig. 1). Their lengths were in the range 85–125 Å and width 25–40 Å. In addition, long linear chains of these rods were seen, and in some regions individual units within the polymers could be distinguished. Our data indicate that the small elements observed in the “dimer” preparation may be composed of two tRNA molecules associated side-to-side in a parallel manner.

It thus appears that an important feature common to both monomeric and “dimeric” tRNA^{Ser} is the presence of short threadlike structures 80–125 Å in length forming end-to-end aggregates. Another interesting point is the flexibility of these elements as shown by the presence of both straight rods and V-shaped structures [7, 8]. The number of V-shaped particles is reduced in the “dimeric” preparation as compared to the monomeric one.

A conformational model should explain the tendency of the tRNA molecule to form end-to-end aggregates. This requirement is not fulfilled by present models as they leave only few possibilities for inter-molecular base-pairing.

The cloverleaf arrangement is capable of explaining many physical and chemical observations made on tRNA. We feel, therefore, that a conformational model of end-to-end aggregates of tRNA molecules should accommodate the essential features of the cloverleaf arrangement.

Our data are compatible with a model where the stem of the cloverleaf is opened up and the 3'- and the 5'-ends of the polynucleotide chain separated leading to a segmented molecule composed of alternating double- and single-stranded regions. With this arrangement end-to-end associations between tRNA molecules would be favoured as the 3'-end of one molecule could form base-pairs with the 5'-end of another molecule. This resembles the polymerization of virus nucleic acids with cohesive ends. Zachau [9] has also suggested this mechanism of aggregation between molecules with similar sequences. In single molecules the ends of the polynucleotide strand

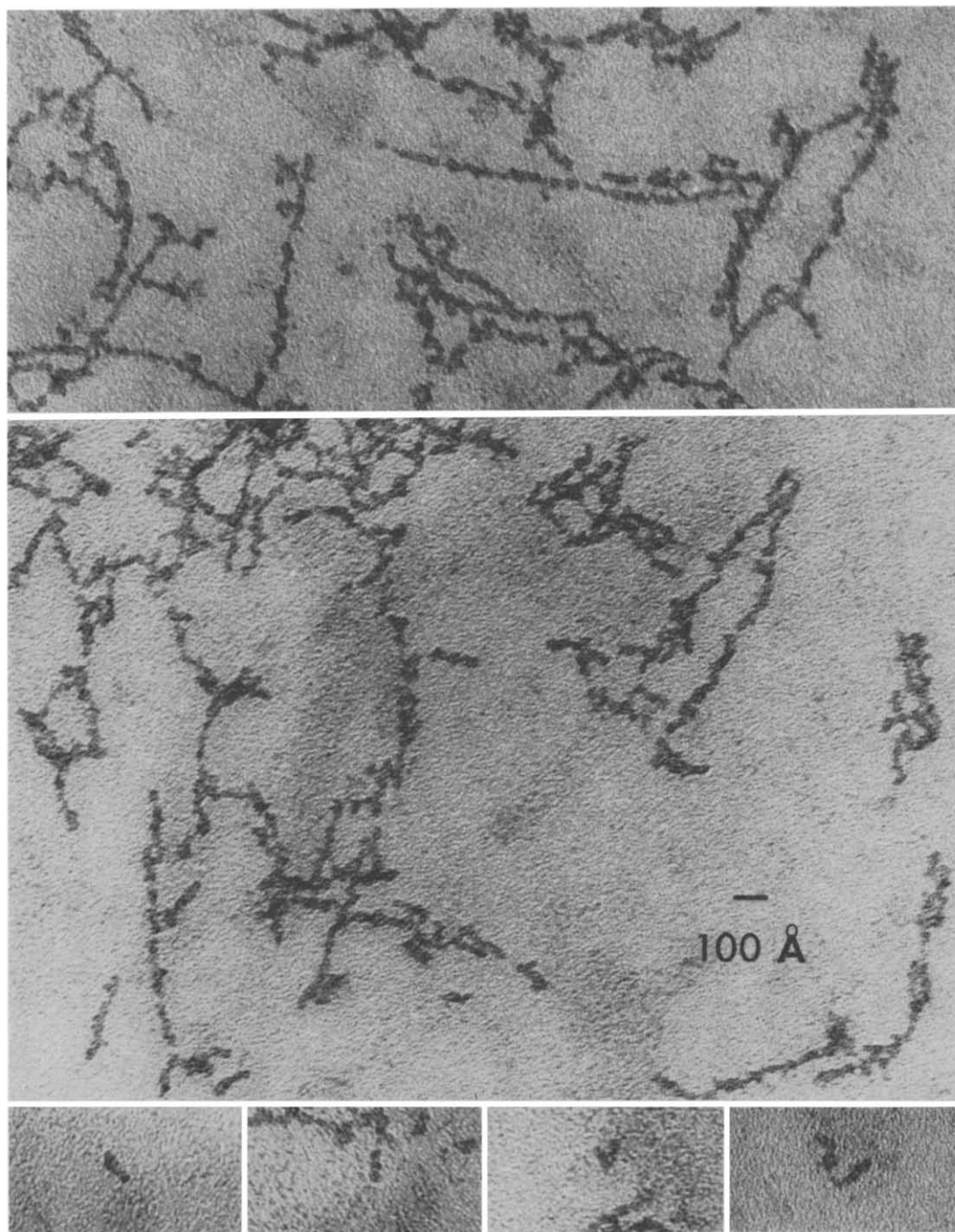


Fig. 1. Positive staining with 1% (w/v) aqueous uranyl acetate of a tRNA^{Ser} fraction which on Sephadex behaves as dimers [9]. The tRNA (9 optical density units per ml) was dissolved in standard cacodylate buffer [9] of pH 7.0. The specimens were prepared on copper grids with thin carbon films. The primary magnification was 100 000 \times , and the total magnification 500 000 \times . Note the presence of long threadlike structures showing some degree of branching. In some places short rods, about 100 Å in length, are observed. A few of these, selected from different micrographs, are shown in the lower part of figure. 1.

might form part of a triple-helical structure with neighbouring helical regions (cf. [6, 10]). Such a model would allow conformational flexibility due to the single strands connecting helical regions. As a consequence, individual molecules could also form more tightly packed structures than would be allowed by the standard cloverleaf arrangement.

Recently, Fuller [11] has proposed a new model for the three dimensional structure of transfer RNA based on the cloverleaf arrangement. According to his view the molecule is 100 Å in length and 20 Å in width at the ends — with a thicker region in the middle. The model also allows some flexibility near the middle and could thus be in agreement with our micrographs. However, end-to-end aggregation does not seem to be favored any more than with the standard cloverleaf arrangement.

Finally it should be stressed that the present model is partly derived from micrographs of tRNA preparations obtained in lyophilized form and dissolved at high concentrations in deionized water or dilute buffer [7, 8]. This could conceivably give rise to artifacts and the structures observed may, therefore, not be identical to the physiologically active ones. Only continued studies can resolve this question.

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